

## REVIEW ARTICLE

## NASAL CAVITY: A PROMISING TRANSMUCOSAL PLATFORM FOR DRUG DELIVERY AND RESEARCH APPROACHES FROM NASAL TO BRAIN TARGETTING

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## ABSTRACT

Drug delivery through nasal route has attracted the interest of scientific community as it has been potentially explored as an alternative route for the administration of vaccines and biomolecules such as proteins, peptides and non-peptide drugs that are susceptible to enzymatic or acidic degradation and first-pass hepatic metabolism. The nasal mucosa is one of the most permeable and highly vascularized site for drug administration ensuring rapid absorption and onset of therapeutic action. In addition it minimizes the lag time associated with oral drug delivery and offers noninvasiveness, self medication, patient comfort and patient compliance which are hurdled in intravenous drug therapy. The objective of this review is to provide an anatomical, histological and physiological overview of nose, absorption enhancers, barriers related to nasal drug delivery, physicochemical, biological and formulation related factors affecting nasal drug delivery system and its advantages. It also highlights research approaches on brain targeting through nasal cavity.

**Keywords:** - Intranasal drug delivery, Absorption enhancer, Barriers to intranasal delivery, Bioavailability, Brain targeting, Vaccines and biomolecules.

## 1. INTRODUCTION-

Currently, Nasal drug delivery has been recognized as a very promising route for delivery of therapeutic compounds including biopharmaceuticals<sup>1</sup>. Nasal administration is a logical choice for topical nasal treatments such as antihistamines and corticosteroids. The nasal mucosa has also received attention as a viable means of systemic administration of analgesics, sedatives, hormones, cardiovascular drugs, and vaccines.<sup>2</sup> Conventionally, the nasal route has been used for local delivery of drugs for treating nasal allergy, nasal congestion, or nasal infections. However systemic delivery through the nasal route has recently begun to explore possibilities for those requiring a rapid onset of action or necessitating avoidance of severe proteolysis involved in oral administration (e.g., most peptide and protein drugs). Successful attempts to deliver corticosteroid hormones through the nasal route for systemic absorption<sup>3</sup> have triggered further studies in this area.

The anatomy and physiology of the nasal passage indicate that nasal administration has potential practical advantages for the introduction of therapeutic drugs into the systemic circulation. The concentration-time profiles achieved after nasal administration are often similar to those after intravenous administration, resulting in a rapid onset of pharmacological activity<sup>4</sup>. Bjerre *et al.* showed that the sedative propiomazine, for which a rapid onset of action is desirable, and it absorbed within 5 minutes after nasal administration to rats.<sup>5</sup> Drugs ranging from small chemicals to large macromolecules including peptide/protein therapeutics, hormones, and vaccines, are being delivered through the nasal cavity. Marketed products include a range of antimigraine drugs (e.g., sumatriptan, zolmitriptan, ergotamine, and butorphanol) as well as some peptides (e.g., calcitonin, desmopressin, and buserelin).<sup>6</sup> Later the use of the nasal route for delivery of vaccines, especially against respiratory infections such as influenza, is attracting interest from vaccine delivery scientists.<sup>7</sup>

Currently, nasal administration is used therapeutically for the systemic absorption of drugs in a variety of indications, including sumatriptan for migraine<sup>8</sup>, the antidiuretic desmopressin for the treatment of diabetes insipidus<sup>9</sup> and oxytocin for secretion of milk in response to suckling during breast feeding or contraction of the uterine muscle to hasten childbirth by nasal delivery. Other drugs still in the research and development pipeline, which have potential for administration nasally, include vitamin B12,<sup>10</sup> various benzodiazepines<sup>11-12</sup> and the dopamine agonist apomorphine for patients with Parkinsonism.<sup>13</sup> Nicotine is also available in a nasal dosage form for use in assisting smoking cessation

1.1 Advantages of Nasal Drug Delivery Systems<sup>14-15</sup>

- Large nasal mucosal surface area for dose absorption
- Rapid drug absorption via highly vascularized mucosa
- Rapid onset of action
- Ease of administration, noninvasive
- By pass the BBB
- Avoidance of the gastrointestinal tract and first pass metabolism
- Improved bioavailability
- Direct transport into systemic circulation and CNS is possible
- Lower dose/reduced side effects
- Improved convenience and compliance
- Self-administration

## 1.2 Limitations-

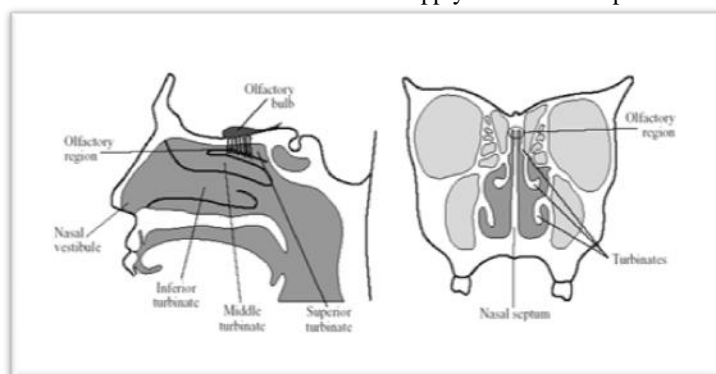
- Volume that can be delivered into nasal cavity is restricted to 25-200  $\mu$ l.
- Not feasible for high molecular weight more than 1k Da
- Adversely affected by pathological conditions
- Drug permeability may alter due to ciliary movement

- e) Drug permeability is limited due to enzymatic inhibition
- f) Nasal irritants drugs cannot be administered through this route
- g) Exact mechanism is not yet clearly known

## 2. ANATOMY AND PHYSIOLOGY OF NOSE

The human nose is divided by the median septum, a central partition of bone and cartilage; each symmetrical half opens at the face via the nostrils and connects with the mouth at the nasopharynx. The nasal vestibule, the respiratory region and the olfactory region are the three main regions of the nasal cavity.<sup>16</sup> The lateral walls of the

nasal cavity include a folded structure which enlarges the surface area in the nose to about 150 cm<sup>2</sup>.<sup>17</sup> This folded structure includes three turbinates. The superior, the median and the inferior. In the main nasal airway, the passages are narrow, normally only 1–3 mm wide and this narrow structure enables the nose to carry out its main functions.<sup>18</sup> During inspiration, the air comes into close contact with the nasal mucosa and particles such as dust and bacteria are trapped in the mucus. Additionally, the inhaled air is warmed and moistened as it passes over the mucosa. This conditioning of the inhaled air is facilitated by the fluid secreted by the mucosa and the high blood supply in the nasal epithelium.<sup>17-18</sup>

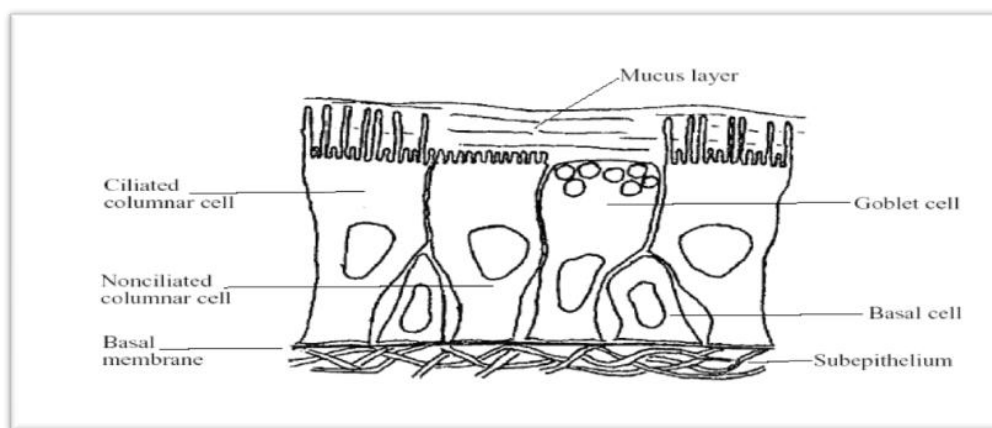


**Figure 1:** Anatomy of the nose (To the right is a cross-section of the nose)

The sub mucosal zone of the nasal passage is extremely vascular and this network of veins drains blood from the nasal mucosa directly to the systemic circulation, thus avoiding first-pass metabolism<sup>19</sup> the nasal cavity is covered with a mucous membrane which can be divided into non-olfactory and olfactory epithelium areas. The non-olfactory area includes the nasal vestibule, which is lined with skin-like cells and respiratory region.

### 2.1 The Respiratory Region<sup>20-25</sup>

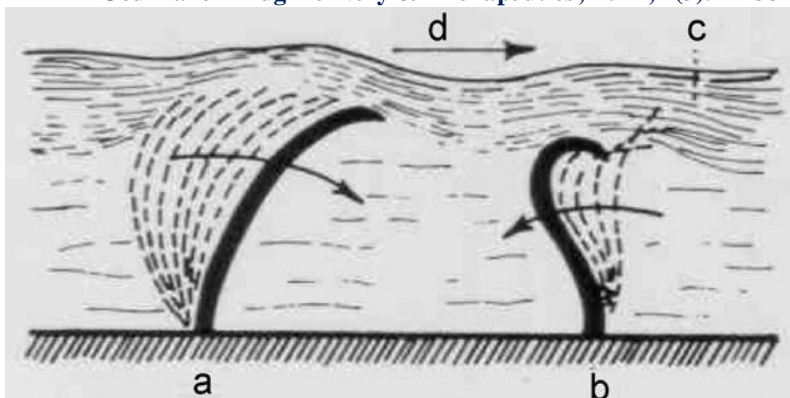
The respiratory epithelium is composed of four types of cells, namely, non-ciliated and ciliated columnar cells, basal cells and goblet cells. These cells facilitate active transport processes such as the exchange of water and ions between cells and motility of cilia (where applicable). They may also serve to prevent drying of the mucosa by trapping moisture.



**Figure 2:** Respiratory epithelium of the nasal cavity, showing the four main types of cells

About 15–20% of the respiratory cells are covered with a layer of long cilia, which move in a coordinated way to propel mucus towards the pharynx. Mucus (or nasal secretion) is a complex mixture of materials, consisting of approximately 95% water, 2% mucin, 1% salts, 1% of other proteins such as albumin, immunoglobulins, lysozymes and lactoferrin and <1% lipids (Kaliner et al., 1984). Mucus is present in two layers on the epithelium in

order to facilitate mucociliary clearance. A viscous gel layer, the 'mucus blanket' (Fig. 3c; 'gel layer', 2–4 mm thickness) floats on the serous fluid layer (Fig. 3e 'sol layer', 3–5 mm thickness). The viscous gel layer is moved along by the hook shaped cilia termini during the energy dependent 'effective stroke' phase of the ciliary motion (Fig. 3).



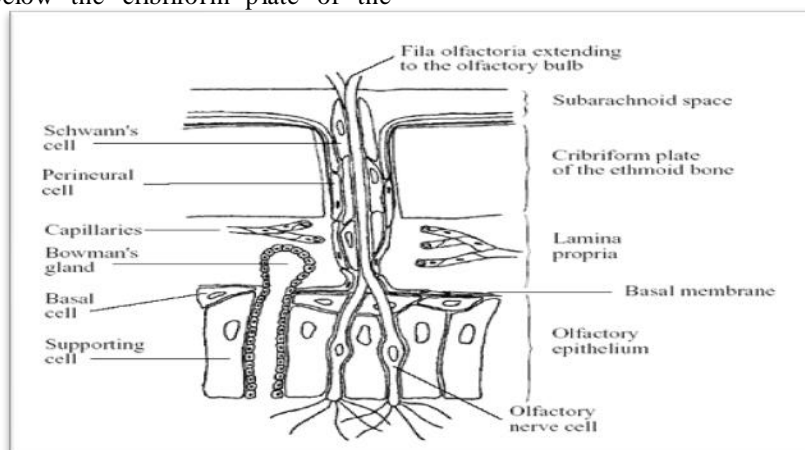
**Figure 3:** The relationship between ciliary motion and mucus layer composition that allows mucociliary clearance. (a) Effective Stroke, (B) Recovery Stroke, (C) Gel Layer, (D) Direction of Gel Layer Movement, (E) Sol Layer.

Cilia are up to 7mm in length when fully extended but can fold to half this length during the recovery stroke where the hook terminus detaches from the gel layer and moves immersed in the sol layer in the opposite direction to the gel layer movement (Fig. 3b). The cilia beat with a frequency of 1000 strokes per min. Hence the mucus moves only in one direction from the anterior to the posterior part of the nasal cavity to the nasopharynx

## 2.2 The Olfactory Region [20-25]

In human, the olfactory region is located on the roof of the nasal cavities, just below the cribriform plate of the

ethmoid bone, which separates the nasal cavities from the cranial Cavity. The olfactory tissue is often yellow in color, in contrast to the surrounding pink tissue. Humans have relatively simple noses, since the primary function is breathing, while other mammals have more complex noses better adapted for the function of olfaction.<sup>24</sup> The olfactory epithelial layer predominantly contains three cell types: the olfactory neural cells, the sustentacular (also known as supporting) cells and the basal cells. Basal cells are progenitor cells (of supporting cells) that also provide mechanical support via anchorage to other cells.(Fig-4)



**Figure 4:** Anatomical connections between the olfactory mucosa in the nose and the CSF in the subarachnoid space outside the olfactory bulb.

The olfactory epithelium is a gateway for substance entering the CNS and peripheral circulation. The neuronal connection between the nasal mucosa and brain provide a unique pathway for the non-invasive delivery of therapeutic agents to CNS. The olfactory neuronal pathway provides both intraneuronal and extraneuronal pathways into the brain. The intraneuronal pathway involves axonal transport and require to hours to days for drugs to reach different brain regions. While the extraneuronal pathway probably relies on the bulk flow transport through perineural channels, which delivers drugs directly to the brain parenchymal tissue or CSF. The extraneuronal pathway allows therapeutic agents to reach the CNS within minutes.

Thorne et al.<sup>25</sup> have reported that the trigeminal neural pathway may also be involved in rapidly delivering protein therapeutic agents, such as insulin like growth factor-1 to brain and spinal cord following intranasal administration.

The transport of drugs across the nasal membrane and into the bloodstream may involve either passive diffusion of drug molecules through the pores in nasal mucosa, Including blood supply, nerve supply or some form of non passive transport.

## 3. BARRIERS FOR NASAL DRUG DELIVERY

### 3.1 Low bioavailability

Bioavailability of polar drugs is generally low, about 10% for low molecular weight drugs and not above 1% for peptides such as calcitonin and insulin.<sup>26</sup> The most important factor limiting the nasal absorption of polar drugs and especially large molecular weight polar drugs such as peptides and proteins are the low membrane permeability. Drugs can cross the epithelial cell membrane either by the transcellular route exploiting simple concentration gradients by the receptor mediated or vesicular transport mechanisms or by the paracellular route

through the tight junctions between the cells. Polar drugs with molecular weights below 1000 Da will generally pass the membrane using the latter route.<sup>27</sup> Although tight junctions are dynamic structures and can open and close to a certain degree when needed, the mean size of these channels is of the order of less than 10 Å and the transport of larger molecules is considerably more limited. Larger peptides and proteins are able to pass the nasal membrane using an endocytotic transport process but only in low amounts.<sup>28</sup> Nasal absorption of such polar drugs can be greatly improved by co administration of absorption enhancing agents.<sup>29</sup> Agents generally used for transnasal absorption includes surfactants (laureth-9, sodium laurylsulfate), bile salts ,bile salt derivatives (sodium glycocholate, sodium deoxycholate, sodium taurodihydrofusidate), fatty acids,fatty acid derivatives (linoleic acid), phospholipids (lysophosphatidylcholine), various cyclodextrins and cationic compounds like chitosan and its derivatives, poly-L-arginine, poly-L-lysine.<sup>30</sup> These enhancers work by a variety of mechanisms but generally they act by altering the permeability of the epithelial cell layer by modifying the phospholipid bilayers, leaching of proteins from the membrane or even stripping off the outer layer of the mucosa. Some of these enhancers also have an effect on the tight junctions and /or work as enzymatic degradation inhibitors. With such absorption enhancing agents, increased bioavailability was obtained, even for larger peptides such as insulin. In animal studies it has been shown for a range of enhancing agents that there is a direct correlation between the absorption enhancing effect and the damage to the nasal mucosa.<sup>31</sup> This is particularly true for bile salts and surfactants. For other enhancers, such as cyclodextrins and chitosan the enhancing effect outweighs the damage caused to the mucosa. Hence, it is of great importance to consider the choice of absorption enhancer for a nasally delivered drug that is not easily absorbed, especially in terms of potential nasal and systemic toxicity.

### 3.2 Mucociliary clearance

The general fast clearance of the administered formulation from the nasal cavity due to the mucociliary clearance mechanism is another factor of importance for low membrane transport. This is especially the case when the drug is not absorbed rapidly enough across the nasal mucosa. It has been shown that for both liquid and powder formulations, which are not bioadhesive, the half life for clearance is of the order of 15 - 30 min.<sup>32,33</sup> The use of bioadhesive excipients in the formulations is an approach to overcome the rapid mucociliary clearance. The clearance may also be reduced by depositing the formulation in the anterior, less ciliated part of the nasal cavity thus leading to improved absorption.<sup>34,35</sup>

### 3.3 Enzymatic Degradation-

Another contributing, but often less considered factor to the low bioavailability of peptides and proteins across the nasal mucosa is the possibility of an enzymatic degradation of the molecule in the lumen of the nasal cavity or during passage through the epithelial barrier. These sites both contain exo-peptidases such as mono and diaminopeptidases that can cleave peptides at their N and C terminals and endo-peptidases such as serine and cysteine, which can attack internal peptide bonds.<sup>36</sup> The

use of enzyme inhibitor may be approaches to overcome this barrier.<sup>37</sup> In summary, the nose offers unique advantages as administration site for drug delivery. However, low permeability for polar and high molecular weight drugs, rapid clearance of the delivery system from the cavity and possible enzymatic degradation of the drug in the nose may be encountered. These challenges can be faced by various approaches, such as use of bio-adhesive systems and absorption enhancers.

## 4. MECHANISM OF DRUG ABSORPTION THROUGH NOSE

The first step in the absorption of drug from the nasal cavity is passage through the mucus. Small unchanged particles easily pass through this layer. However, large or charged particles may find it more difficult to cross. Subsequent to a drug's passage through the mucus, there are several mechanisms for absorption through the mucosa. These include transcellular or simple diffusion across the membrane, paracellular transport *via* movement between cell and transcytosis by vesicle carriers.<sup>33</sup> Obstacles to drug absorption are potential metabolism before reaching the systemic circulation and limited residence time in the cavity. Several mechanisms have been proposed but the following two mechanisms have been considered predominantly.

- The first mechanism involves an aqueous route of transport, which is also known as the paracellular route. This route is slow and passive. There is an inverse log-log correlation between intranasal absorption and the molecular weight of water-soluble compounds. Poor bio-availability was observed for drugs with a molecular weight greater than 1000 Daltons.<sup>38</sup>
- The second mechanism involves transport through a lipoidal route that is also known as the transcellular process and is responsible for the transport of lipophilic drugs that show a rate dependency on their lipophilicity. Drugs also cross cell membranes by an active transport route *via* carrier-mediated means or transport through the opening of tight junctions.<sup>38</sup> For example, Chitosan, a natural biopolymer opens tight junctions between epithelial cells to facilitate drug transport.<sup>39</sup>

## 5. CRUCIAL FACTORS FOR NASAL FORMULATIONS<sup>40</sup>

The deposition of drug and deposition area is mainly dependant on the delivery system and the delivery device because of nasal cavity has the peculiar anatomy and physiology. The deposition is influenced by factors such as mode of administration, particle size of the formulation, velocity of the delivered particles, spray angle and cone. A wider spray cone could result in loss of the formulation while narrower spray cones would lead to limited deposition with respect to the available absorption surface area. An optimal spray cone may effectively and rapidly deliver the drug from the formulation at desired specific site/s present within nasal cavity. The selection of delivery system depends upon the drug used, therapeutic indication, patient population, and last but not least, marketing preferences. In addition to dosage form design, pharmacokinetics and bioavailability of drugs may be governed by several factors following intranasal



administration. Some of the physicochemical, formulation, and physiological factors must be considered prior to designing an intranasal delivery system for CNS targeting.

## PHYSICOCHEMICAL PROPERTIES OF DRUGS -

Physicochemical properties are one of the important aspects in design of nasal formulation

### 6. FACTORS RELATED TO DRUG

#### 6.1 Lipophilicity

Absorption of drug substance through biological membrane may be dependent on hydrophilic lipophilic balance of the compound.<sup>41</sup> On increasing lipophilicity, the nasal absorption of the compound normally increases.<sup>42</sup> Although In one study it was found that lipophilic compounds alprenolol and propranolol were well absorbed from the nasal mucosa, in comparison to the hydrophilic drug metoprolol. Lipophilic compounds tend to readily cross biological membranes via the transcellular route since they are able to partition into the lipid (bilayer) of the cell membrane and diffuse into and traverse the cell in the cell cytoplasm. Nasal absorption of steroids was directly correlated with lipophilicity of drug molecules and was found to be independent of pH.<sup>43</sup> A number of lipophilic drugs such as naloxone, buprenorphine, testosterone<sup>44</sup> and 17 $\alpha$ -ethinyloestradiol,<sup>45</sup> have been shown to be completely or almost completely absorbed nasally in animal models.

#### 6.2 Partition Coefficient and pKa

According to the pH partition theory, unionized form of drug are well absorbed compared with ionized form of drug and the same theory is applicable in the case of nasal drug absorption. Jiang et al.<sup>46</sup> conducted a study to determine the quantitative relationship between the physicochemical properties of drugs and their nasal absorption, using diltiazem hydrochloride and paracetamol as model drugs. The results showed that a quantitative relationship existed between the partition coefficient and the nasal absorption constant.

#### 6.3 Chemical form

The chemical form of a drug is important in determining absorption. For example, conversion of the drug into a salt or ester form can alter its absorption. Huang et al.<sup>47</sup> reported that in-situ nasal absorption of carboxylic acid esters of L-tyrosine was significantly greater than that of unmodified L-tyrosine.

#### 6.4 Molecular weight

A linear inverse correlation has been reported between the absorption of drugs and molecular weight up to 300 Daltons. Absorption decreases significantly if the molecular weight is greater than 1,000 Daltons except with the use of absorption enhancers. Based on the reports by Fisher et al.<sup>48</sup> and Yamamoto et al.<sup>49</sup> it can be concluded that the permeation of drugs less than 300 Da is not significantly influenced by the physicochemical properties of the drug like molecular weight, size, formulation pH, pKa of molecule, which will mostly permeate through aqueous channels of the biological membrane.

#### 6.5 Particle size

It has been reported that particles greater than 10  $\mu$ m in size are deposited in the nasal cavity. Particles that are 2 to

10  $\mu$ m can be retained in the lungs, and particles of less than 1  $\mu$ m are exhaled.

### 6.6 Solubility and dissolution rate

Drug solubility and dissolution rates are important biopharmaceutical factors<sup>50</sup> in determining nasal absorption from powders and suspensions. The particles deposited in the nasal cavity need to dissolve prior to absorption. Solubility of a drug or dosage form is the first prerequisite for absorption and bioavailability of dosage form. The fluid available for dissolution of drug particles in nasal cavity or mucosa is very less when compared to the gastrointestinal fluid in oral drug delivery.<sup>51</sup> saturation solubility of drug in a given nasal physiological pH is very important parameter, which determines the rate and extent absorption of nasal dosage form.

## 7. FORMULATION FACTORS

### 7.1 pH of formulation

Both the pH of the nasal cavity and pKa of a particular drug need to be considered to rationalize systemic absorption. Nasal irritation is minimized when products are delivered with a pH ranging between 4.5 and 6.5.<sup>52</sup> Lysozyme is found in nasal secretions, which is responsible for destroying certain bacteria at acidic pH.<sup>53</sup> Under alkaline conditions, lysozyme is inactivated and the nasal tissue is susceptible to microbial infection. It is therefore advisable to keep the formulation at a pH of 4.5 to 6.5 keeping in mind the physicochemical properties of the drug that absorbed in the un-ionized form. The delivery volume is limited by the size of the nasal cavity. An upper limit of 25 mg/dose and a volume of 0.1-0.2 ml/nostril have been suggested.

The pH of a nasal formulation is important for the following reasons:

- To avoid irritation of nasal mucosa
- To allow the drug to be available in unionized form for absorption
- To prevent growth of pathogenic bacteria in the nasal passage
- To maintain functionality of excipients such as preservatives
- To sustain normal physiological ciliary movement

### 7.2 Osmolarity

Drug absorption can be affected by tonicity of the formulation. Ohwaki et al.<sup>43</sup> studied the effect of osmolarity on the absorption of secretin in rats and found that absorption reached a maximum at a sodium chloride concentration of 0.462 M, because shrinkage of the nasal epithelial mucosa was observed at in the presence of hypertonic solutions.<sup>54</sup> Hypertonic saline solutions are also known to inhibit or cease ciliary activity. Low pH has a similar effect on cells as hypertonic solutions.

### 7.3 Gelling agents

Retention of the nasal formulation in the nasal cavity can enhance therapeutic effect by virtue of enhancing rate and extent of drug absorption. According to a study by pennington et al.<sup>55</sup> increasing the viscosity may provide a means of prolonging the effect of nasal formulation. Suzuki, et al.<sup>56</sup> showed that a drug carrier, hydroxyl-propylcellulose was effective for improving the absorption

of low molecular weight drugs but did not produce the same effect for high molecular weight peptides.

#### 7.4 Solubilizers

The aqueous solubility of a drug is always a limitation for nasal drug delivery in solution. Conventional solvents or co-solvents such as glycols, small quantities of alcohol, Transcutol (diethylene glycol monoethyl ether), medium chain glycerides and Labrasol (saturated polyglycolized C8-C10 glyceride) can be used to enhance the solubility of drugs. Other options include the use of surfactants or cyclodextrins such as hydroxypropyl-beta-cyclodextrin that serve as biocompatible solubilizers and stabilizers in combination with lipophilic absorption enhancers. In such cases, impact of the solubilizers on nasal irritancy should be considered.

#### 7.5 Drug concentration, required dose, and dose volume

Drug concentration, dose and volume of administration are three interrelated parameters that impact the performance of the nasal delivery system. Ex vivo experiments in rats demonstrated the effect of drug concentration on nasal drug absorption. Nasal absorption of I-tyrosyl-L-tyrosine was found to increase with increasing concentration of drug. However few experiments showed different effects of drug concentration on the absorption of drugs, for example the absorption of aminopyrine from rat nasal mucosa was constant as a function of its concentration.<sup>57</sup> Interestingly, nasal absorption of salicylic acid was decreased with increasing concentration of administered drug and low absorption of high concentration of salicylic acid was lined with its nasal epithelial toxicity and nasal membrane resistance.<sup>58</sup>

### 8. ROLE OF ABSORPTION ENHANCERS

The selection of absorption enhancers is based upon their acceptability by regulatory agencies and their impact on nasal physiological function. Absorption enhancers may be required when a drug exhibits poor membrane permeability, large molecular size, lack of lipophilicity and susceptibility to enzymatic degradation by aminopeptidases.

Generally, the absorption enhancers act via one of the following mechanisms:

- Inhibit enzyme activity
- Reduce mucus viscosity or elasticity
- Decrease mucociliary clearance
- Open tight junctions; and
- Solubilize or stabilize the drug

Absorption enhancers are generally classified as physical and chemical enhancers. Chemical enhancers act by destructing the nasal mucosa very often in an irreversible way, whereas physical enhancers affect nasal clearance reversibly by forming a gel. The enhancing effect continues until the gel is swallowed. Examples of chemical enhancers are chelating agents, fatty acids, bile acid salts, surfactants, and preservatives. Osmolarity and pH may accelerate the enhancing effect.<sup>48</sup>

### 9. PHYSIOLOGICAL FACTORS

#### 9.1 Effect of deposition on absorption

Deposition of the formulation in the anterior portion of the nose provides a longer nasal residence time and better absorption.<sup>59</sup> The dosage form deposited in posterior chamber of nasal cavity will be eliminated by nasal mucociliary clearance and hence show low bioavailability. The site of deposition and deposition pattern of liquid dosage form is dependent on the delivery device, mode of administration, physicochemical properties of drug molecule.<sup>60</sup>

Harris<sup>61</sup> compared the deposition and removal of metered dose sprays with nasal drops. Nasal sprays were deposited anteriorly, after which small portions were cleared slowly into nasal pharynx by mucociliary clearance. In contrast, drops were deposited mostly posteriorly and were removed rapidly in large portions into the nasal pharynx.

#### 9.2 Nasal blood flow

The nasal mucosal membrane is very rich in vasculature and plays a vital role in the thermal regulation and humidification of inhaled air.<sup>18,19</sup> Drug absorption will depend upon the vasoconstriction and vasodilatation of these blood vessels.<sup>24</sup>

#### 9.3 Effect of mucociliary clearance

It is important that the integrity of the nasal clearance mechanism is maintained to perform normal physiological functions such as the removal of dust, allergens and bacteria. The absorption of drugs is influenced by the residence time between the drug and the epithelial tissue. The mucociliary clearance is inversely related to the residence time and therefore inversely proportional to the absorption of drugs administered. A prolonged residence time in the nasal cavity may also be achieved by using bioadhesive polymers,<sup>34</sup> microspheres, chitosan, and polycarbophil, or by increasing the viscosity of the formulation.

#### 9.4 Effect of enzymatic activity

Several enzymes that are present in the nasal mucosa might affect the stability of drugs. For example, proteins and peptides are subjected to degradation by proteases and amino-peptidase at the mucosal membrane.<sup>37</sup>

#### 9.5 Effect of pathological condition

Intranasal pathologies such as allergic rhinitis, infections, or previous nasal surgery may affect the nasal mucociliary transport process and/or capacity for nasal absorption. During the common cold, the efficiency of an intranasal medication is often compromised. Nasal clearance is also reduced in insulin-dependent diabetes. Nasal pathology can also alter mucosal pH and thus affect absorption of drugs. An increased level of albumin and plasma proteins in nasal fluid was found in allergic rhinitis.<sup>62</sup> However in recent study by Person [63] and coworkers has clearly shown that intranasal absorption of macromolecules is not increased allergic rhinitis. However introduction of nasal dosage form as an external stimulus increases the nasal mucus secretion and leads to drainage dosage forms and hence reduced bioavailability.<sup>64</sup>

### 10. NASAL FORMULATIONS

The deposition and deposition area are mainly a function of the delivery system and delivery device. Different dosage forms and their application to deliver the drugs to

the central nervous system following intranasal drug delivery are discussed in this section.

## 10.1 LIQUID DOSAGE FORMS

Liquid dosage forms either in form of soluble, suspended or colloidal systems are normally used for formulating nasal delivery systems.

### 10.1.1 Nasal drops

Nasal drops are one of the most simple and convenient delivery systems among all formulations. The main disadvantage of this system is the lack of dose precision.<sup>65</sup> It has been reported that nasal drops deposit human serum albumin in the nostrils more efficiently than nasal sprays.

### 10.1.2 Nasal sprays

Both solution and suspension formulations can be formulated into nasal sprays. Due to the availability of metered dose pumps and actuators, a nasal spray can deliver an exact dose anywhere from 25 to 200  $\mu$ L. The particle size and morphology (for suspensions) of the drug and viscosity of the formulation determine the choice of pump and actuator assembly. Solution and suspension sprays are preferred over powder sprays because powder results in mucosal irritation.<sup>66</sup>

### Nasal emulsions, microemulsions and nanoparticles

Intranasal emulsions and nanoparticles have not been studied as extensively as other liquid nasal delivery systems. Nasal emulsions offer the advantages for local application mainly due to the viscosity. One of the major disadvantages is poor patient acceptability. The physical stability of emulsion formulations and precise delivery are some of the main formulation issues.

## 10.2 SEMI-SOLID DOSAGE FORMS

Semi-solid systems, for example gels, ointments and liquid systems containing polymers that gel at particular pH changes are usually employed for designing the nasal drug delivery systems.

### 10.2.1 Nasal gels

Nasal gels are thickened solutions or suspensions, of high-viscosity. The advantages of a nasal gel include the reduction of post-nasal dripping due to its high viscosity, reduction of the taste impact due to reduced swallowing, reduction of anterior leakage of the formulation, reduction of irritation by using soothing/emollient excipients, and target delivery to the mucosa for better absorption.<sup>67</sup> Vitamin B12 and apomorphine gel have been successfully employed to achieve desired therapeutic concentrations following nasal administration.

## 10.3 SOLID DOSAGE FORMS

Solid dosage forms are also becoming popular for intranasal drug delivery, although these formulations are more suitable for pulmonary drug delivery and similar applications, since it can cover the vasculature within the epithelium of nasal mucosa.

### 10.3.1 Nasal powders

Powder dosage forms may be developed if solution and suspension dosage forms cannot be developed, mainly due

to lack of drug stability. The advantages of a nasal powder dosage form are the absence of preservative and superior stability of the drug in the formulation. However, the suitability of the powder formulation is dependent on the solubility, particle size, aerodynamic properties and nasal irritancy of the active drug and/or excipients. An additional advantage of this system is local application of drug, but nasal mucosa irritancy and metered dose delivery are some of the challenges for formulation scientists and device manufacturers who are interested in powder dosage forms.<sup>38</sup>

## 11. APPLICATION OF NASAL DRUG DELIVERY SYSTEM

### 11.1 Delivery of Vaccines through Nasal Route

Nasal delivery of vaccines has been reported to not only produce systemic immune response, but also local immune response in the nasal lining, providing additional barrier of protection. The majority of the invading pathogens enter the body *via* mucosal surfaces.<sup>68</sup> Therefore, mucosal sites have a potential as first line of defense against entering pathogens. Pathogens are filtered from the inspired air by compaction and mucociliary clearance. Nasal secretions are known to contain immunoglobulins (IgA, IgG, IgM, IgE), protective proteins such as complement as well as neutrophils and lymphocytes in the mucosa.<sup>69</sup>

Delivering the vaccine to the nasal cavity itself stimulates the production of local secretory IgA antibodies as well as IgG, providing an additional first line of defense system.<sup>69</sup>

- Main reasons for exploiting the nasal route for vaccine delivery
- The nasal passages are rich in lymphoid tissue.
- Creation of both mucosal and systemic immune responses.
- Low cost, patient compliance, non-injectable, and safe.

The feasibility of the nasal route for administering vaccines against plague, diphtheria tetanus,<sup>70</sup> influenza,<sup>71</sup> cholera,<sup>72</sup> and HIV<sup>73</sup> has already been tested for inducing both mucosal and systemic immune response against the carried antigen. Read et al<sup>74</sup> have prepared nasal influenza vaccine using chitosan. Nasal influenza vaccination may prove to be a good alternative to parenteral injection because of the enhancement of the mucosal immune response and the ease of vaccine administration. This study investigated the use of chitosan, a bioadhesive polymer, as a nasal delivery system with inactivated, subunit influenza vaccine. Nagamoto et al.<sup>75</sup> prepared a novel vaccine carrier particulate system (nano-particles and emulsions) with chitosan and had evaluated the effect of this system on the immune response for intranasal delivery. Chitosan nanoparticles (NP) and chitosan-coated emulsions (CC Emul) were prepared by improvement of the method previously reported and by modified ethanol injection methods, respectively. Jiang et al.<sup>76</sup> prepared chitosan microspheres by ionic gelation process with sodium sulfate for nasal vaccine delivery. Bordetella Bronchiseptica Dermonecrototoxin (BBD) as a major virulence factor of a causative agent of atrophic rhinitis (AR) was loaded to the chitosan microspheres for vaccination.

**Table 1: Nasal Drug Product for Vaccination Available in the Market**<sup>71,74,76</sup>

Vaccine (Product name)	Dosage form	Status	Manufacturer
Human influenza vaccine (Nasalflu Berna)	Virosomes (Spray)	Marketed (withdrawn)	Berna Biotech
Equine influenza vaccine (Flu Avert)	Drops	Marketed	Heska
Porcine Bordetella bronchiseptica vaccine (Maxi/Guard Nasal Vac)	Drops	Marketed	AddisonBiological Laboratory
Feline Bordetella bronchiseptica vaccine (Nobivac Bp)	Suspension drops	Marketed	Intervet
Human Streptococcus A vaccine (StrepAvax)	Proteosomes (nanoparticulate)	Phase 2	ID Biomedical
Human influenza vaccine (FluInsuru)	Proteosomes (nanoparticulate)	Phase 2	ID Biomedical
Human influenza vaccine	Not indicated.	Phase 1/2	West PS
Human influenza vaccine (FluMist)	Spray	Marketed	MedImmune Inc.
Feline trivalent vaccine against calici herpes-1 and parvovirus	Drops	Marketed	Heska

### 11.2 Nose to Brain drug delivery system

Delivery of drugs to the central nervous system (CNS) remains a challenge in the development of efficacious agents for central targets, mainly due to the impenetrable nature of the blood-brain barrier (BBB). The utility of the nasal route as a portal for preferential delivery of therapeutic agents is in the focus of scientists. The BBB limits substrate penetration based on several characteristics, including lipophilicity, molecular size and specificity for a variety of ATP-dependent transport systems.<sup>1</sup>

At the beginning of the previous century the direct connection between the brain and the open air was discovered: the olfactory neurons. Injection of dyes in the ventricles of rabbits and monkeys showed that the cerebrospinal fluid (CSF) is drained via the olfactory neurons into the olfactory neurons, originating from the olfactory bulb, connect the brain with the nasal cavity by penetrating the cribriform plate, which brings the neurons into the nasal mucosa. This coined the idea that this transport route could also exist in the opposite direction, which would imply direct access from the nasal cavity to the brain, thus circumventing the BBB. This opened up a new perspective in the research field of drug targeting to the CNS, lymphatic vessels and the nasal mucosa. A growing numbers of recent reports have demonstrated the effectiveness of intranasal administration of neuroprotective agents in decreasing ischemic brain injury. For example, Ying et al.<sup>77</sup> recently reported that intranasal administration of NAD<sup>+</sup> profoundly decreased brain injury in a rat model of transient focal ischemia.

Similarly, Wei et al.<sup>77</sup> showed that intranasal administration of the PARG inhibitor Gallo tannin decreased ischemic brain injury in rats. Such agents are believed to provide neuroprotection by diminishing or abolishing activation of poly (ADP-ribose) polymerase-1 (PARP-1), which plays a significant role in ischemic brain damage. NAD<sup>+</sup> was observed to reduce infarct formation by up to 86% even when administered at 2 hours after ischemic onset.

Recently, Shi et al.<sup>78</sup> Investigated the extent of systemic absorption and uptake of meptazinol (MEP) hydrochloride in cerebrospinal fluid (CSF) after intranasal administration on rats and compared with oral administration. CSF samples were collected by a serial sampling method. The concentration of MEP in the biological samples was measured by HPLC with

fluorescence detector. A rapid and significant level of MEP in the plasma and CSF was achieved after nasal administration whereas the oral administration resulted in considerably lower drug concentrations. The area under curve (AUC) in plasma and CSF from the nasal route was 7.375 and 15.6 folds compared with those of the oral route, respectively. The outcome of the research indicated that intranasal MEP is able to show quick absorption and improve the bioavailability, which could be a promising alternative to oral administration..

Chen et al.<sup>79</sup> investigated the potential of delivering nerve growth factor to the brain along the olfactory neural pathway for the treatment of Alzheimer's disease and found that the nerve growth factor reached the brain within an hour achieving the highest concentration in the olfactory bulb and less in other brain regions. Hui Yu et al.<sup>80</sup> developed direct nose-to-brain delivery of a growth hormone releasing neuropeptide, hexarelin after intranasal administration to rabbits and investigate the olfactory transfer of a growth hormone releasing neuro-peptide, hexarelin to the brain tissues by comparing brain uptake levels after intranasal administration with those after intravenous administration. Frey declared that by nasally administering insulin-like growth factor (IGF-1) the drug could bypass the blood brain barrier and reach the central nervous system directly from the nasal cavity.<sup>81</sup>

Tomotaka Shingakia et al.<sup>82</sup> The transnasal delivery of 5-fluorouracil to the rat brain is enhanced by acetazolamide and evaluate the effect of acetazolamide (AZA), an inhibitor of the secretion of cerebrospinal fluid (CSF), on the direct drug transport from the nasal cavity to the CSF and the brain uptake of a model drug, 5-fluorouracil (5FU). Kao et al.<sup>83</sup> studied the nasal transport of various esters, such as butyl and methyl, of the carboxyl group of L-dopa in the rat model and found significantly higher levels of L-dopa in the CSF and the olfactory bulb than did equimolar doses given intravenously.

Kumar et al.<sup>84</sup> have studied the nasal uptake of tritium-labeled estradiol and progesterone in rhesus monkeys. Both compounds were found to be absorbed intranasally and were able to penetrate into cerebrospinal fluid rapidly. A comparison of the plasma:CSF ratio of the two compounds indicated more steroids accumulated in the CSF after ability and minimizing variation. They demonstrated intranasal administration compared to the intravenous route. This result suggested that the two steroids can be absorbed from the nasal cavity and reach the brain directly via the respiratory and olfactory mucosa.



Sakane et al.<sup>85</sup> reported that following intranasal administration of the antibiotic cephalexin to rats, higher CSF concentration was reached at 15 min but it declined to approximately half that concentration at 30 min. Because cephalexin do not cross the BBB well and CSF concentration was 166-fold higher after intranasal administration than after systemic administration in spite of similar blood levels, it was concluded that cephalexin entered the CSF directly from the nasal cavity. Using a

series of fluoresceine inisothiocyanate-labeled dextrans (FITC-dextran) with increasing molecular weights, it was found that dextrans with molecular weights of upto 20,000 daltons could be transported directly from the nasal cavity of rats into the CSF. The concentration of the FITC-dextrans in the CSF increased with decreasing molecular weight. These FITC-dextrans were not found in the CSF after intravenous administration.

**Table 2: Drugs and drug-related compounds reported to reach the CNS after nasal administration in different animal models**

Drug molecule	Species	Sample	Method	References
Adenoviral lacZ vector	Hamster Mouse	Brain	Histochemical	105
Bupivacaine	Rat	CSF	HPLC	106
Cephalexin	Rat	CSF	HPLC	85
Chlorpheniramine	Rat	CSF	HPLC	107
Cocaine	Rat	Brain tissue	HPLC	108
D4T	Rat	CSF	HPLC	109
Dihydroergotamine	Monkey	Brain tissue	Radioactivity counting	110
Fibroblast growth factor	Mouse	CSF	Motor activity,Dopamine activity	111
Estradiol	Rabbit	CSF	Radioactivity counting	84
Tetracaine	Rat	CSF	HPLC	105
Hydroxyzine	Mouse	CSF	HPLC	112
Insulin	Rat	Brain tissue	Radioactivity counting	113,114
Lidocaine	Rat	CSF	HPLC	115
Midazolam	Dog	CSF	HPLC	116
Monosialoganglioside	Rat	CSF	Immuno-enzymatic assay	117
Nerve growth factor	Rat	Brain tissue CSF	ELISA, Radioactivity counting	79
$\beta$ -Alanine (as carnosine)	Hamster Mouse	Brain tissue/CSF	Autoradiography,Biochemical analysis	118

### 11.3 Delivery of Peptide and Non-Peptide Drugs for Systemic Effect through Nasal Route

Most peptides and proteins, being hydrophilic polar molecules of relatively high molecular weight, are poorly absorbed across biological membranes with bio-availability obtained in the region of 1–2% concentrations when administered as simple solutions.<sup>86</sup> This low uptake may be adequate for the development of some commercial products like desmopressin and calcitonin because they have a wide therapeutic index. But for certain peptide drugs such as insulin which does not have the luxury of wide therapeutic index it is essential to develop the novel formulation strategies.

In order to produce a product with a good bioavailability that can provide sufficient reliability in dosing and overcome these problems much research has been carried out in the areas of absorption enhancers and bioadhesive agents. Absorption enhancers are used to increase the bioavailability, and these enhancers are basically surfactants, glycosides, cyclodextrin and glycols. Recent studies have shown that the high bioavailability achieved with absorption enhancers for the delivery of polar compounds across mucosal membranes can be associated with tissue damage.<sup>87</sup> Hence care should be taken that the absorption enhancer used should not only increase the bioavailability but should also be harmless to the nasal mucosa. The classical example of a polypeptide compound with low nasal bioavailability is calcitonin. Its molecular weight is approximately 3,500 daltons and

contains 32 amino acid in length. when calcitonin was given intranasally to rats and rabbits using a number of different cyclodextrins, its absorption when measured as decrease in serum calcium concentration, was found to be significant in comparison to the formulation without cyclodextrin addition.<sup>88</sup> It has been observed that increasing the time of contact with the nasal mucosa can increase the nasal absorption of insulin. The clearance half life can be increased from 15 min with nasal solution to 240 min using starch microspheres (SMS).Insulin administered in combination with SMS resulted in 497% increase in AUC for plasma insulin as compared to insulin solution. The AUC increased by 1657% compared to insulin solution when an enhancer lysophosphatidylcholine was used with insulin and SMS [70]. When a surfactant such as saponin, sodium glycolate or BL-9 was added to the preparation, the absorption of insulin from the nasal mucosa was enhanced independent of pH.<sup>89</sup> Absorption enhancing effect of different cyclodextrins on intranasally administered insulin in rats and rabbits was determined. Dimethyl-beta-cyclodextrin was found to be most potent.<sup>90,91</sup> Nasal absorption is also promoted by medium chain fatty acid salt<sup>92</sup> glycyrrhetic acid derivatives in rat<sup>93</sup> sodium-tauro-24, 25-dihydrofusidate in sheep.<sup>94</sup> Etr et al.<sup>95</sup> demonstrated that the blood glucose remained low for 3 h after lunch when insulin was used intranasally with 1% deoxycholate in type1 diabetics. Other proteins like luteinizing hormone releasing hormone,<sup>96</sup> growth hormone<sup>97</sup> and adreno-corticotrophic hormone<sup>98</sup> have been administered intranasally.

**Table 3: Nasal Drug Products (Proteins and Peptides) for Systemic Drug Delivery** <sup>(2,3,6,7)</sup>

Drug Substance (Product name)	Indication	Dosage form	Status	Manufacturer
Salmon calcitonin (Karil 200 I.E.)	Osteoporosis	Solution (spray)	Marketed	Novartis Pharma
Desmopressin (Minirin Nasenspray)	Antidiuretic hormone	Solution (spray)	Marketed	Ferring Arzneimittel
Buserelin (Profact nasal)	Prostate cancer	Solution (spray)	Marketed	Aventis Pharma
Nafarelin (Synarel)	Endometriosis	Solution (spray)	Marketed	Pharmacia
Oxytocin (Syntocinon)	Lactation induction	Solution (spray)	Marketed	Novartis Pharma
Protirelin (antepan* nasal) (Relefact* TRH nasal)	Thyroid diagnostics	Solution (spray)	Marketed	Sanofi-synthelabo Aventis Pharma

**Table 4: Nasal Drug Products (Non-Peptide) for Systemic Drug Delivery on the Market** <sup>(2,3,6,7)</sup>

Drug Substance (Product name)	Indication	Dosage form	Status	Manufacturer
Zolmitriptan (AscoTop* Nasal)	Migraine	Solution (spray)	Marketed	Astra Zeneca
Sumatriptan (Imigran* Nasal)	Migraine	Solution (spray)	Marketed	Glaxo SmithKline
Dihydroergotamin (Migranal* Nasal Spray)	Migraine	Solution (spray)	Marketed	Novartis Pharma
Estradiol (Aerodiol*)	Hormone replacement	Solution (spray)	Marketed	Servier

Unlike high molecular weight peptides, the small non-peptide lipophilic drugs (MW below 1000) are better absorbed through the nasal mucosa even in the absence of absorption enhancers. The underlying epithelium of the nasal membrane is highly vascularized and the nasal cavity has a large surface area readily accessible for drug absorption because of the presence of nasal turbinates. As a consequence, low molecular weight lipophilic drugs, such as propranolol,<sup>99</sup> progesterone<sup>100</sup> are well absorbed across the nasal cavity and resulting in a faster onset of action.<sup>86</sup>

Recently, Chavan et al.<sup>101</sup> have tried to increase the nasal absorption of sumatriptan succinate by using bile salts. A rat in situ nasal perfusion technique was used to examine the rate and extent of absorption of sumatriptan succinate. In vitro enzymatic drug degradation studies were carried out with rat nasal washings. Various experimental conditions such as nasal perfusion rate, pH of the perfusion medium and concentrations of absorption enhancers such as sodium deoxycholate, sodium caprate, sodium tauroglycocholate and EDTA were optimized. In vivo studies were carried out for the optimized formulation in rabbits and the pharmacokinetics parameters of nasal solution were compared with marketed nasal solutions. Nasal absorption of sumatriptan succinate was pH dependent. It was found maximum at pH 5.5 and decreased at higher pH values. In in-vitro enzymatic degradation studies, no measurable degradation was observed during the first week. The extent of drug absorption was increased by absorption enhancers. Sodium deoxycholate appeared to be more effective for enhancing the nasal absorption of sumatriptan succinate than the other absorption enhancers. The order of increasing absorption of sumatriptan succinate caused by the enhancers was sodium deoxycholate > sodium caprate > sodium tauroglycocholate > EDTA.

Wermeling et al.<sup>102</sup> have evaluated the efficacy and tolerability of a sterile, unpreserved butorphanol tartrate nasal spray administered via a unit-dose device in the treatment of moderate to severe pain after dental impaction surgery. In this study, sterile nasal spray of butorphanol

tartrate administered via a unit-dose device provided effective post surgical analgesia in approximately half of patients who had under-gone surgery to remove impacted third molars. Intranasal route has also been tried with limited success for drugs such as steroids (corticosteroids, estradiol, testosterone, and so on),<sup>103, 104</sup> anti-hypertensives (nifedipine, nitroglycerine, propranolol, hydralazine), analgesics (buprenorphine, morphine), antibiotics and antivirals.<sup>106-109</sup>

## 12. CONCLUSION –

Drug delivery through nasal route provides a practical, non invasive method of by passing the blood brain barrier (BBB) in order to deliver therapeutic agents to the brain. This method allows drugs that do not cross the BBB to be delivered to the central nervous system in a few minutes along with both the olfactory and trigeminal neuronal pathway. This delivery system has clinical benefits like reduction in drug dosage and systemic exposure, which results in lesser side effects. It is expected that nasal drug formulations will continue reach to the market and this delivery will include not only drugs for acute and chronic disease but also novel nasal vaccines for local or systemic protection against microbial infection. However it is well known that intranasal route has several limitations which must be overcome to develop a successful nasal formulation. Drug related factor and pathophysiological condition of nose determine the nasal drug absorption. Some common approaches, like increasing the nasal residence time of drug, use of absorption or penetration enhancers and minimization of the mucociliary clearance, enhances the bioavailability of nasally administered drug. This route is believed to be an alternative route to oral and parenteral because of the successful administration of vaccines and biomolecules such as proteins, peptides and non-peptide drugs, that are susceptible to enzymatic or acidic degradation and first-pass hepatic metabolism. Moreover it also offers noninvasiveness, self medication, patient comfort and patient compliance which are hurdled in intravenous drug therapy.

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